**Structure *16***

## Supplemental Data

# Bacteriophage Lambda Stabilization by Auxiliary Protein gpD: Timing, Location, and Mechanism of Attachment Determined by Cryo-EM

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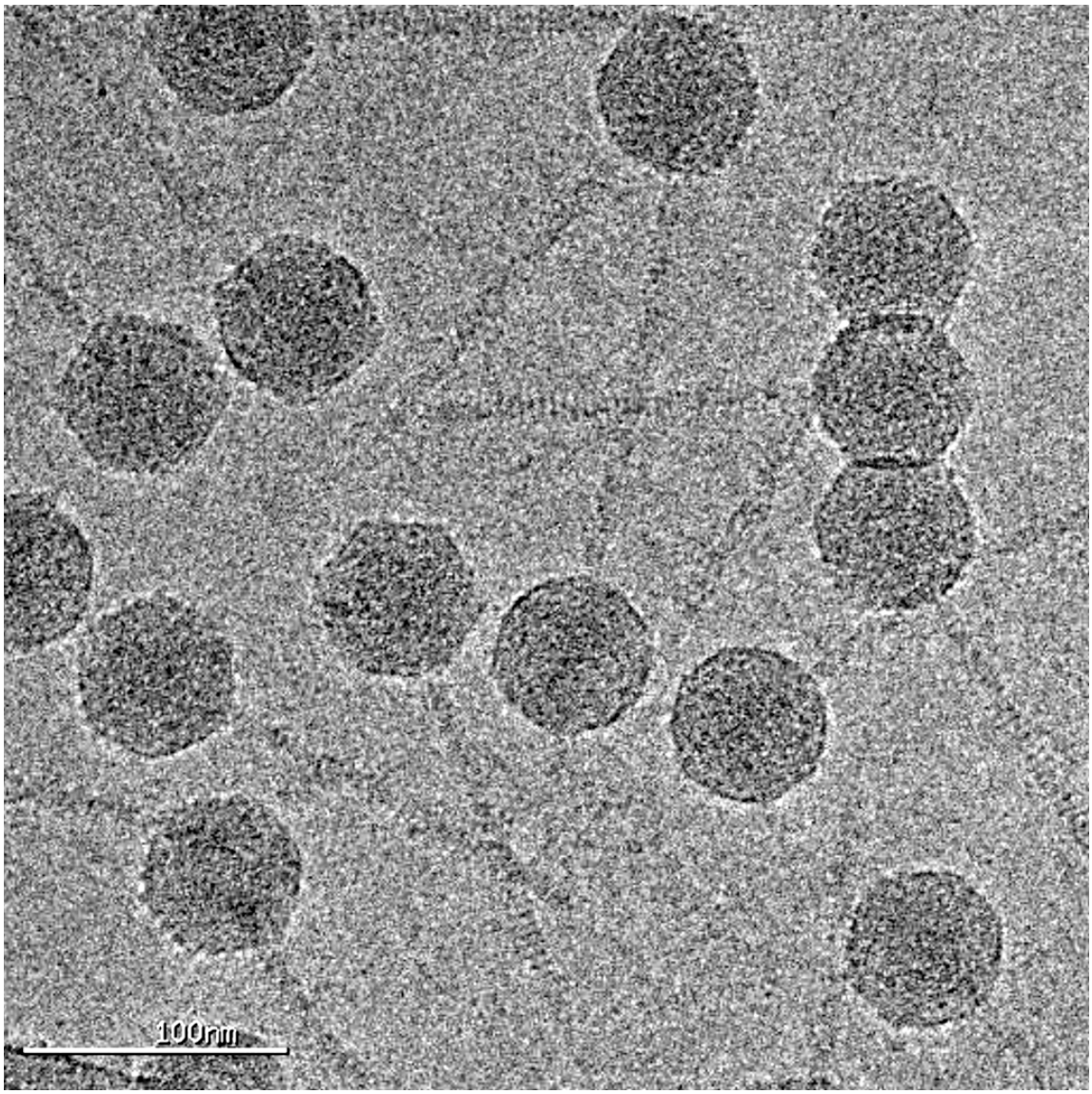


**Figure S1. A CryoEM Image of Bacteriophage Lambda Embedded in Vitreous Ice, Taken at 200-kV at Liquid Nitrogen Temperatures on a Tietz 4Kx4K CCD Camera**
The DNA-filled icosahedral capsids and long filamentous tails are clearly seen in this representative image.

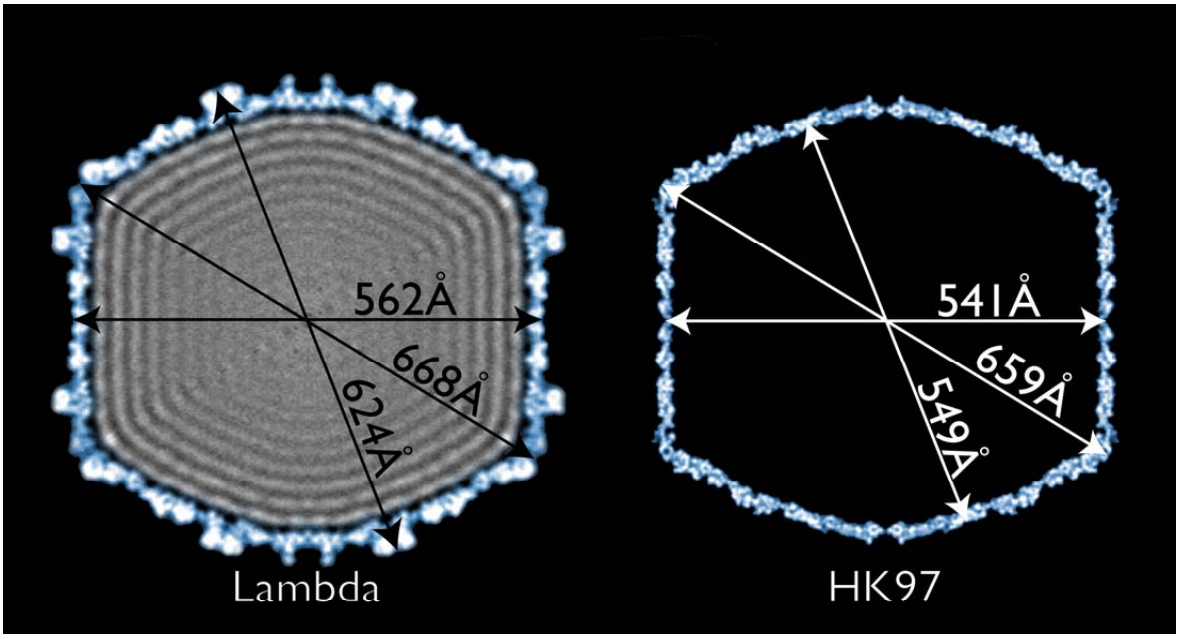


**Figure S2. Comparison of the Lambda and HK97 Capsid Dimensions**
Central slices of the lambda reconstruction exhibit similar dimensions to those of the HK97 crystal structure. The only substantial difference is along the three-fold axis, where phage lambda is an additional 75Å in diameter due to the attachment of gpD. The concentrically packaged genome is also clearly visible within the capsid interior at a spacing of 24Å from one DNA strand center to the next.

```
         1    .    10     .    20     .    30     .    40     .    50     .
         TSKETFTHYQPQGNSDPAHTATAPGGLSAKAPAMTPLMLDTSSRKLVAWDGTTDGAA
psipred:--EEE------------EEEEE------------EEE------EEE--------E
samdssp:--EEEE------TTS--EEEEEEEETTE---------EE-TTTSEEEEEES--TS-E
sstride:--EEEETT---TTTT--TEEEEETTTT--EEET--EEEEETTTTEEEEETTTTTTEE
jnet:   --EEEEE--E--------EEEE----------------------EEE--------E

          60     .    70     .    80     .    90     .   100
         VGILAVAADQTSTTLTFYKSGTFRYEDVLWPEAASDETKKRTAFAGTAISIV
         EEEEEEEE----EEEEEEEE-EEEEEEE-----HHHHHHHHHHH----EEE-
         EEEEEEEESSS-EEEEEEEEEEEEEEEE--GG--HHHHHHHHHH--EEEE-
         EEEEEEE-----EEEEEEEE-EEEEEEE------HHHHHHHH-------EE-
         EEEEEEEETTTT-EEEEEEEEEEEEEEEETTTTH-HHHHHHHHHH-------
```

**Figure S3. Secondary Structure Prediction of gpD Sequence by Four Different Servers (Cuff et al., 1998; Frishman and Argos, 1995; Karplus et al., 2003; McGuffin et al., 2000)**.
The sequence is colored based on the secondary structure seen in the crystal structure, with the first fourteen disordered residues colored in green. It is evident that the predictions are generally in agreement with the experimental data, such that a notion of a beta strand at the N-terminal end of the disordered region is well-founded.

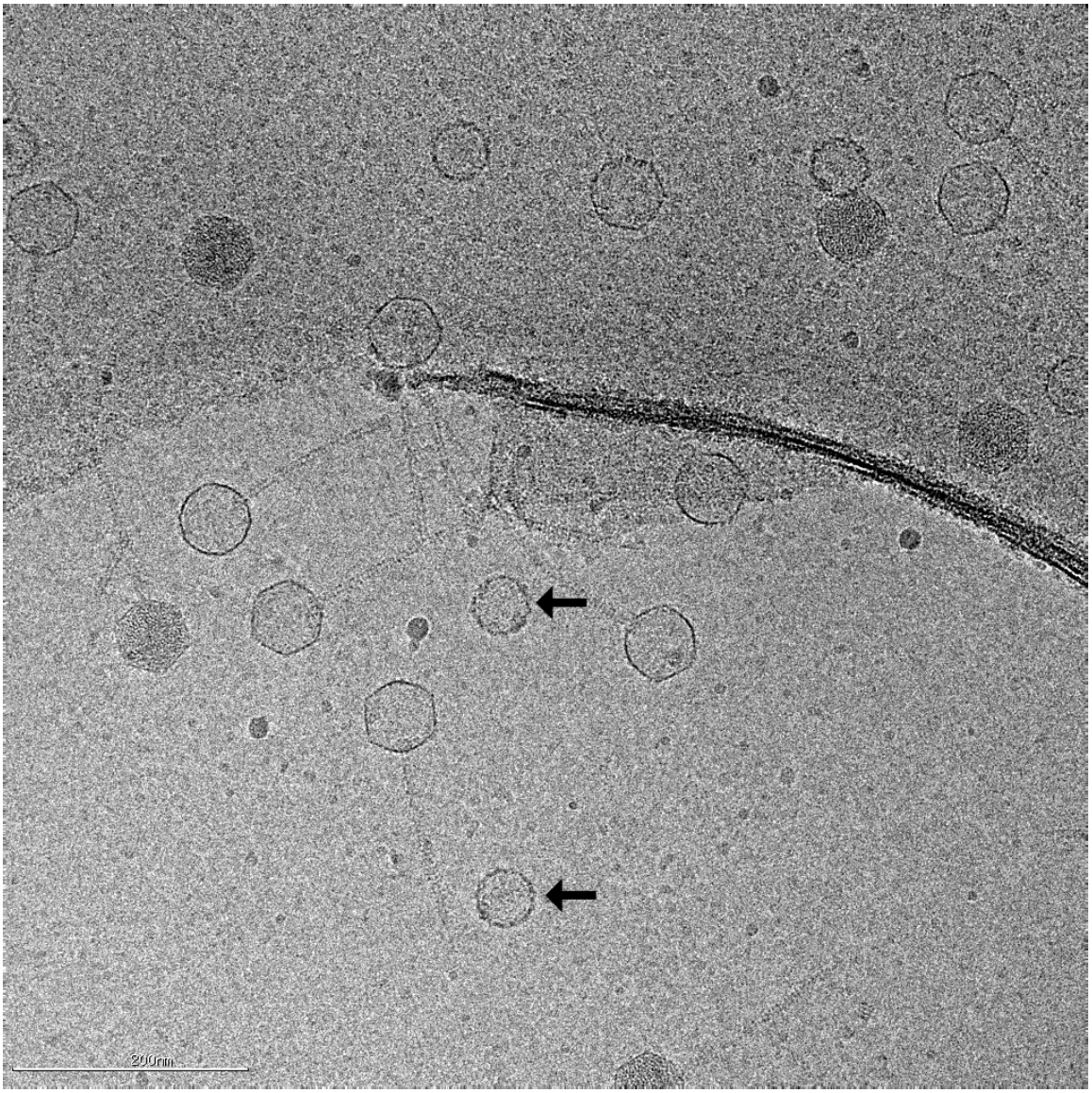


**Figure 4. A CryoEM Image of 37.7 kbp-Packaging Mutant Bacteriophage Lambda Embedded in Vitreous Ice, Taken at 200-kV at Liquid Nitrogen Temperatures on a Gatan 4Kx4K CCD Camera**

Two prohead particles, identified by black arrows, are smaller, rounder, and thicker shelled than their mature counterparts in this representative image.

## Supplemental References

Cuff, J.A., Clamp, M.E., Siddiqui, A.S., Finlay, M., and Barton, G.J. (1998). JPred: a consensus secondary structure prediction server. Bioinformatics. *14*, 892–893.

Frishman, D., and Argos, P. (1995). Knowledge-based protein secondary structure assignment. Proteins. *23*, 566–579.

Karplus, K., Karchin, R., Draper, J., Casper, J., Mandel-Gutfreund, Y., Diekhans, M., and Hughey, R. (2003). Combining local-structure, fold-recognition, and new fold methods for protein structure prediction. Proteins. *53*, 491-496.

McGuffin, L.J., Bryson, K., and Jones, D.T. (2000). The PSIPRED protein structure prediction server. Bioinformatics.*16*, 404–405.